Pathogenicity, Host Response and Control of Cercospora Leaf-Spot of Sugar Beets

By Edgar F. Vestal

AGRICULTURAL EXPERIMENT STATION
IOWA STATE COLLEGE OF AGRICULTURE
AND MECHANIC ARTS

R. E. Buchanan, Director

BOTANY AND PLANT PATHOLOGY SECTION

RESEARCH BULLETIN NO. 168
December, 1933
AMES, IOWA

December, 1933 Research Bulletin No. 168

Pathogenicity, Host Response and Control of Cercospora Leaf-Spot of Sugar Beets

By Edgar F. Vestal

AGRICULTURAL EXPERIMENT STATION
IOWA STATE COLLEGE OF AGRICULTURE
AND MECHANIC ARTS

R. E. Buchanan, Director

BOTANY AND PLANT PATHOLOGY SECTION

AMES, IOWA

CONTENTS

Page

Summary 43

Growth habits and sporulation of *Cercospora beticola* on artificial media 45

Development of conidia and conidiophores . . 46
Germination of conidia on the leaves of a living plant 47
The influence of changing humidity upon penetration of stomata by Cercospora mycelia . . . 48

Infection trials on sugar beet seedlings . . . 49

The host range of *Cercospora beticola* 50

Inoculation experiments with *Cercospora beticola* . 51
Beta vulgaris inoculations with *Cercospora beticola* 51
Results of cross inoculations 56
Cercospora beticola found in the field . . . 57

Control of Cercospora leaf-spot 61

Spraying and dusting for Cercospora leaf-spot . . 61
Spraying and dusting in 1929 62
Spraying and dusting in 1930 63

The influence of spacing upon the development of Cercospora leaf-spot 64
Field trial in 1931 65
Artificial inoculation 66
Development of Cercospora leaf-spot in the drilled and checked beets 66
Comparison of environmental factors . . . 69
Literature cited 70

SUMMARY

On artificial media conidia of *Cercospora beticola* began to appear in 12 to 20 hours after inoculation, and the optimum production was from 48 to 96 hours after transfer.

Germination of the conidium may take place at any point, but more often near a septum and from the basal end of the cell first.

Germination of the conidia did not take place on the living leaf in an atmosphere containing less than 90 percent humidity.

The humidity of the leaf surface was found to be an average of 3.5 percent higher than the surrounding atmosphere.

An air humidity of 75 percent was necessary for any conidial formation, and increasing humidity from 75 to 96 percent produced an increase in conidial length from 100.4μ to 185.2μ.

Only a small percentage of the germ tubes growing over the surface of the leaf penetrated the stomata.

Varying the humidity of the atmosphere surrounding the plant did not appear to influence the penetration of the stomata by the germination tubes.

Cercospora beticola readily infected seedlings of sugar beet, producing both hypocotyl and cotyledon infection.

Twenty-six species of plants, distributed throughout 12 widely separated families were infected under greenhouse conditions by spraying with suspensions of conidia of *C. beticola.* The finding of 26 plants which could be infected with *C. beticola* clearly indicates that the organism has a wide host range among the plants of the sugar beet area.

Under field conditions six species of plants in six widely separated families have been found infected with a *Cercospora sp.* morphologically and pathogenically similar to *C. beticola.*

Cercospora beticola was found growing in the field under conditions which indicated that it might at times be a saprophyte.

The application of fungicides for the control of Cercospora leaf-spot did not produce a significant increase in the yield of beets and sugar.

Statistical analysis of returns from machine blocking indicate significantly greater yields of beets from 20-inch blocking than from either 12 or 24-inch. The percentage of sugar was slightly greater in the beets grown in the 12-inch blocking, but it was not significant; the increase in tonnage of the 20-inch blocking more than compensated for the difference in the percentage of sugar. No significant differences were found in purity.

Blocking the sugar beets 20 inches in the row retarded the leaf-spot spread about 10 days as compared with the 12-inch blocking. This retardation of leaf-spot development among the 20-inch blocking reduced the number of dead leaves as much as 16 percent below that of the 12-inch blocking.

The use of machinery makes possible a much more rapid and uniform cultivation than is usually given by hand. This makes possible, in turn, a much more effective control of the weeds and a maintenance of a lower humidity among the beet leaves. Thus the same measures which reduce the spread of leaf-spot will also reduce the cost of production and increase the yield in 20-inch blocking.

Pathogenicity, Host Response and Control of Cercospora Leaf-Spot of Sugar Beets*

By Edgar F. Vestal**

Cercospora leaf-spot (*Cercospora beticola* Sacc.) of sugar beets presents one of the most serious problems confronting the sugar beet industry. By destruction of the leaves during the growing season, this organism reduces not only the weight of beets, but also the purity and percentage of sugar. Nearly every sugar beet section of the world has suffered from this disease. In this country the loss in many sections is frequently 50 percent of the crop. No beet growing section has escaped entirely, although the irrigated districts of Utah, Idaho and California have suffered less than those of the more humid areas. Under climatic conditions such as those of Iowa and neighboring states to the north and west, the disease becomes a yearly menace to the sugar beet industry and probably is one of the important limiting factors in this section of the United States.

Progress in the direction of control has been slow, due to the lack of adequate knowledge of the conditions which initiate an epidemic of the pathogene, especially the source of primary infection. In this study attention was first focused upon the possibility of weed hosts as a source of primary infection. It was soon apparent, however, that more information about the growth habits and the pathogenicity of the organism was necessary before much progress on host range could be made.

GROWTH HABITS AND SPORULATION OF *CERCOSPORA BETICOLA* ON ARTIFICIAL MEDIA

Before the pathogenicity and host range of *Cercospora beticola* could be determined, it was necessary to be able to sporulate the organism at will. Pool and McKay (34) reported that they were unable to secure conidia of *C. beticola* in pure culture. Duggar (16) had previously reported that he was unable to secure conidia of *C. beticola* on artificial media when he employed methods which were successful with *C. apii*. Wenzel (50) was unable to produce conidia in quantity when experimenting with plant tissue, various vitamins, methyl and ethyl alcohol and alternation of temperatures; but he secured weak production of

* Taken from a thesis submitted to the Graduate Faculty of Iowa State College in partial fulfillment of the requirements for the degree doctor of philosophy. Project No. 75 of the Iowa Agricultural Experiment Station.

** The writer wishes to express his sincere thanks and appreciation to Dr. I. E. Melhus and Dr. S. M. Dietz for their kindly advice and assistance during the pursuit of this problem and in the preparation of the manuscript, and to Dr. J. C. Gilman and Dr. J. E. Sass for many helpful suggestions.

conidia by the use of a chlorophyll solution in a grape sugar medium. Schmidt (41) cultured *C. beticola* on 2 percent extract of sugar beet leaves and found that the most conidia were obtained when the temperature was alternated between 25° and 15°C. Growth was also obtained on various sterile leaf tissues, such as beet, nettle, spinach and Fuchsia, but conidial production was not mentioned. Coons and Larmer (9) grew *C. beticola* well on standard media, but beet leaf agar proved to be the most satisfactory medium. Conidia were produced over the entire colony, but in greater numbers along the margins. Nagel (31), using a special technique, reported that he grew *C. beticola, C. davisii* Ell. & Ev., *C. dubia* (Riess) Wint., *C. zebrina* Pass. and *C. setariae* Atk. on potato dextrose and sugar beet leaf extract agar with abundant conidial production. The conidia appeared on the cultures within 36 to 72 hours after either isolation or transfer and continued to be produced for about 48 hours at a maximum rate, after which the production rate decreased rapidly. When transfers were made every 3 or 4 days, sporulation was continued at a comparatively high rate for an indefinite period of time.

These studies with the growth response and sporulation began with the isolation of the pathogene from an infected sugar beet leaf obtained at Kanawha, Iowa, in the fall of 1931. On artificial media *C. beticola* formed the typical slow growing, dense mat of mycelium. The submerged mycelium consisted of the short, swollen olive-green cells, while the aerial mycelium was slow growing, dark gray at first and later light gray. The conidiophores were an olive-green with a shade of brown and were borne from one to several in a cluster over the mat. They measured 4-6μ x 20-70μ, with from 1 to 3 septations. The conidia were colorless, measuring 1.5-6μ x 30-165μ, with from 2 to 16 septations.

In pure culture, the first conidia began to appear within 12 to 20 hours after either an isolation or a transfer. The optimum production took place at from 48 to 96 hours after transfer. After 4 days, the number of conidia decreased rapidly until, after 12 days, there were about one-fifth as many produced as at the end of the fourth day.

The production of conidia abundantly and at will was one of the most progressive steps in the study of the biology of *C. beticola.* It immediately placed in the hands of the investigators a tool useful in every phase of laboratory research dealing with this organism.

DEVELOPMENT OF CONIDIA AND CONIDIOPHORES

The first step in the formation of conidia under normal conditions appears to be the building up of a small sclerotial mass

of hyphae. From this mass, one to several hyphae grow upright and become conidiophores, frequently borne singly if produced on soft decayed tissue, or in groups if the host tissue is firm.

At first the conidiophore was undifferentiated. Later, as it grew, the tip became somewhat lighter in color, the protoplasm less dense and somewhat more granular. One or more septations appeared at the base of the conidiophore, but these were in the dark, dense, protoplasmic portion. The terminal portion elongated and became more transparent. Here and there aggregates of oil droplets appeared. In a short time, at the basal end of this semi-transparent portion, the oil droplets and cytoplasm became more closely aggregated. Soon the latter organized into a granular band extending across the hypha. This band seemed to form from either wall and gradually to extend across the intervening space. At the same time a slight depression appeared in the hyphal wall at its junction with the band. By this time the semi-transparent portion began to assume the typical conidial shape and its limits were clearly defined.

As the cutting off of the conidium from the conidiophore progressed, the protoplasm and oil droplets began to collect in the conidium at more or less regular intervals, usually from the basal end upward. The process of laying down the septations in the conidium began as described for the septation at the base of the conidium. Although the process of septal formation usually began from the basal end of the conidium, it was not unusual to find this reversed. Conidia have been observed in which the septations were formed irregularly. Subsequent conidia were produced on the same conidiophore, the end of the conidiophore elongating in the same manner as the original hypha.

Germination of the conidium followed the same procedure as the formation of a secondary conidium by the conidiophore. One to two germ tubes emerged from any one cell of the conidium.

GERMINATION OF CONIDIA ON THE LEAVES OF A LIVING PLANT

The conidia to be studied were placed in designated areas on the leaves of sugar beet seedlings. The plants were set into open petri dishes under bell jars. Sufficient water was placed in each dish to insure a high humidity within the bell jar. With the jars sealed, humidities of 95-100 percent were maintained and, by regulating the openings, lower humidities were also maintained within a range of 5 percent.

When the plants were removed from the bell jars, the portions of the epidermis upon which the conidia had been placed were removed with a sharp razor and placed upside down upon slides previously coated with Meyer's egg albumin fixative. This fixed the conidia in place so they might be stained. The slides

TABLE I. THE INFLUENCE OF THE HUMIDITY AND THE TIME OF EXPOSURE UPON THE GERMINATION OF THE CONIDIA OF *CERCOSPORA BETICOLA* UPON SUGAR BEET LEAVES.

Hours exposed to each humidity	Percentage humidity at each exposure	Number conidia counted	Number conidia germinated	Percentage germinated
6	96	90	70	77.7
12	92	58	23	39.5
15	96	85	85	100.0
24	75-80	139	1	0.72
24	83-88	167	0	0.0
24	87	30	0	0.0
24	90	99	26	27.2
24	92	99	70	70.7
48	85	96	0	0.0
48	80-88	89	1	1.1
48	90	30	24	80.0
48	90	68	55	81.0

were permitted to dry, flooded with cotton blue in lacto-phenol for 1 minute, washed with distilled water and counter-stained with eosin. The mycelium and conidia absorbed the blue, and the host tissue absorbed the eosin.

The results are arranged in table I according to the number of hours of exposure to a particular humidity. At 96 percent humidity, 100 percent of the conidia counted had germinated. During 12 hours exposure at 92 percent humidity, only 39.5 percent of the conidia counted had germinated, while during 24 hours at the same humidity 70.7 percent had germinated. When the humidity was reduced to 90 percent for 24 hours only 27.2 percent of the conidia counted had germinated, but when the conidia were exposed to 90 percent humidity for 48 hours the percentage germinating increased to 81. Below 90 percent humidity the germination ceased.

THE INFLUENCE OF CHANGING HUMIDITY UPON PENETRATION OF STOMATA BY CERCOSPORA MYCELIA

During the course of the inoculation work with the Cercospora organism the question arose as to why some mycelial threads penetrated the stomata and others passed over them without entering. It had been noted that only a very small percentage of the germination tubes entered a stoma, although many passed directly over during their growth. Figure 1 illustrates the relationship between stoma and germ tubes of conidia of *Cercospora beticola* on beet leaves. Hull (23) found that in the case of *Puccinia sorghi* a very low percentage of the tubes entered a stomatal opening. From observations made to date, it would appear that they enter purely by accident, there being no evident attraction toward the stomatal opening. Of the enormous number of mycelial tubes which spread about, apparently less than 1 percent found their way into the leaf under the conditions of these experiments.

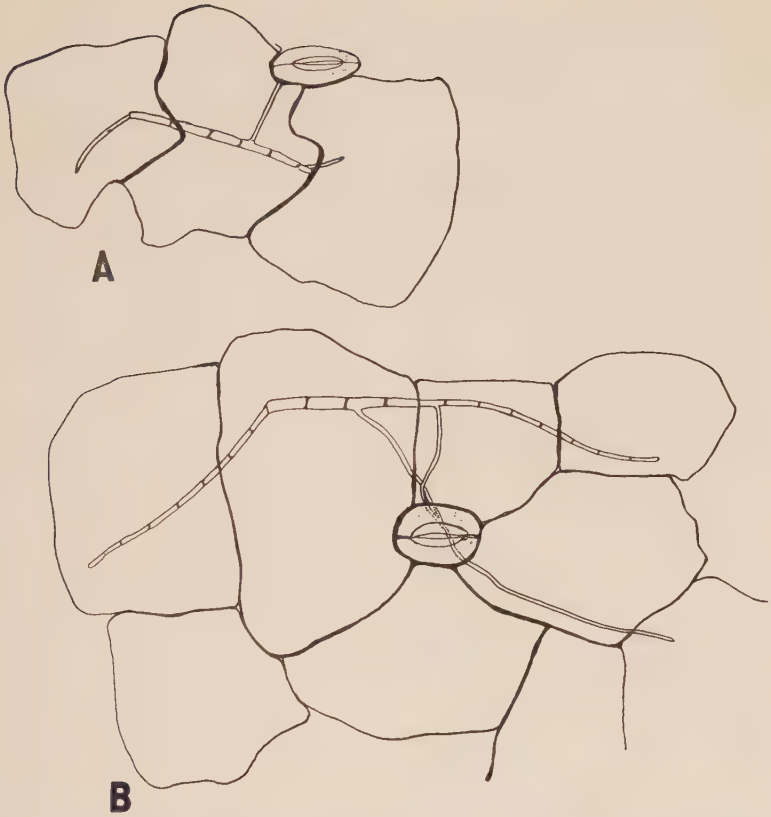

Fig. 1. Relationship between stoma of beet leaf, and germ tube of *Cercospora beticola*: A. Growth of mycelium at the end of 6 hours after spore germination on the leaf; B. growth of mycelium at the end of 15 hours after spore germination on the leaf.

INFECTION TRIALS ON SUGAR BEET SEEDLINGS

The Cercospora leaf-spot disease of sugar beets has generally been considered to be confined to the mature leaves. In 1912 Melhus (28), however, infected mangel seedlings with a suspension of conidia obtained from dead leaves, but his work appears to have been overlooked by Pool and McKay (36) who stated that only the older leaves were infected. Experiments carried on in the greenhouse at Ames, Iowa, during the winter of 1929-30 proved conclusively that seedling infection was easily produced, in fact, more easily produced than infection of mature plants.

During the winter of 1930-31, over 6,000 seedlings in the 2-4 leaf stage were inoculated in the greenhouse, with 100 percent

infection frequently resulting. Inoculation by dusting with powdered infected leaf material or by conidial suspension from pure culture proved equally effective. When plants in the seedling stage were inoculated, a high percentage of cotyledonous infection resulted, with a somewhat smaller percentage showing infection of the hypocotyl (fig. 2). In some cases the infection of the hypocotyl among seedlings grown in flats was so severe as to cause a very perceptible damping off. Suspensions of conidia from diseased sugar beet leaves produced more than twice as many spots per leaf as suspensions of equal spore concentration from pure cultures.

Fig. 2. Infection of cotyledons and hypocotyl of sugar beet seedlings by *Cercospora beticola.*

The humidity of the surface of the leaf, as determined with a precision hygrometer, was found to average 3.55 percent higher than that of the surrounding air. The differences did not appear to be influenced by the percentage of humidity to which the plant was subjected. The average difference of 3.55 percent indicates that there would be sufficient moisture on the leaf surface to germinate conidia when the humidity of the surrounding air would be too low.

Infected sugar beet leaves held at a humidity of 96 percent produced conidiophores and conidia averaging nearly twice the length of those held at 75 percent. Similar differences have been reported by Welles (49) from the Philippines.

THE HOST RANGE OF *CERCOSPORA BETICOLA*

A comparison of the general morphological characters of the species of Cercospora found on the plants common to sugar beet

fields and vicinity disclosed little difference between the character. When *Cercospora beticola* was subjected to various environmental conditions, as discussed in the preceding paragraphs, it showed wider variations in size and form than exist between the species of Cercospora on the plants in the neighborhood of the sugar beets. This variation in the morphology of *C. beticola* introduced the question of species limitations on different hosts. It had already been suggested by Welles (48), Killian (24) and others that morphological characters upon which species classification was based could not differentiate species of Cercospora. *Cercospora beticola,* then, might have a wider host range than was suspected.

Several plants were known to be attacked by *C. beticola.* Pool and McKay (36) reported, that, in addition to *Beta vulgaris,* Swiss chard, mangel wurzel and *Beta vulgaris* var. *cicla,* they were able to infect *Martynia louisiana* Mill. but were unsuccessful with *Amaranthus retroflexus* L., *Chenopodium album* L., *Chenopodium quinoa* L., and *Lactuca sativa* L. Schmidt (41) produced infection on *Atriplex* (Tourn.) L., *Atropa belladonna* L., *Tropaeolum majus* L., and *Urtica dioica* L. He also reported failure with *Chenopodium album.* Nagel (31) secured infection on *Chenopodium album, Rumex acetosella* L., *Rumex crispus* L., *Rumex altissimus* L., and *Spinacea oleracea* L. With this evidence pointing toward a wide range of host plants for *Cercospora beticola,* the problem offered a new and important field for investigation.

INOCULATION EXPERIMENTS WITH *CERCOSPORA BETICOLA*

The work carried on during the winter of 1931-32 was centered about the relationships between *Cercospora beticola* and certain weeds in the sugar beet area. When an inoculation was positive, the fungus was isolated and carried in pure culture. From pure culture it was inoculated on the particular host from which it came, as well as on *Beta vulgaris* and other plants.

BETA VULGARIS INOCULATIONS WITH *CERCOSPORA BETICOLA*

The three varieties of *Beta vulgaris* inoculated appeared to be equally susceptible to *Cercospora beticola* in the trials made with them under greenhouse conditions. The number, size and general character of spots were about the same. The spots on the garden beet were somewhat darker, due to the red in the leaf itself, but otherwise they were very similar. Microscopic measurements of spores isolated from the three varieties of *Beta vulgaris* were uniform.

The garden beet is rather susceptible and is found infected generally throughout the sugar beet area of the state. Beets and Swiss chard were found infected in a garden at Ames, Iowa,

TABLE II. INOCULATION TRIALS ON VARIOUS HOST PLANTS WITH *CERCOSPORA BETICOLA* ISOLATED FROM SUGAR BEETS COLLECTED AT KANAWHA, IOWA, 1931.

Host plants	Number plants inoculated			Total number plants inoculated	Number plants infected	Percentage infection
	Trial 1	Trial 2	Trial 3			
Abutilon theophrasti	4	–	–	4	–	—
Amaranthus retroflexus	5	4	2	11	1	9.09
Apium graveolens	3	2	2	7	5	71.4
Arctium lappa	1	1	4	6	6	100.
Beta vulgaris (Swiss chard)	2	4	3	9	9	100.
Beta vulgaris (red garden beet)	4	4	4	12	12	100.
Chenopodium album	5	5	6	16	11	68.7
Citrullus vulgaris	12	8	4	24	14	58.3
Ipomoea batatis	2	2	2	6	5	83.3
Lactuca sativa	4	1	3	8	4	50.0
Lactuca scariola	2	–	–	2	1	50.0
Malva rotundifolia	2	2	4	8	8	100.
Medicago lupulina	1	1	0	2	–	—
Medicago sativa	1	1	1	3	1?	
Melilotus alba	3	1	–	4	1?	
Pastinaca sativa	1	2	–	3	–	
Plantago lanceolata	3	2	1	6	5	83.3
Plantago major	4	2	2	8	6	75.0
Pisum sativum	1	1	1	3	–	—
Polygonum convolvulus	6	1	1	8	7	87.5
Polygonum pennsylvanicum	4	2	1	7	1	14.2
Portulaca oleracea	1	1	1	3	1	33.3
Rumex acetosella	4	4	3	11	5	45.4
Rumex crispus	5	5	6	16	7	43.7
Setaria glauca	5	–	–	5	–	—
Setaria viridis	5	–	–	5	–	—
Soja max	2	2	2	6	5	83.3
Spinacea oleracea	4	4	4	12	12	100.
Solanum tuberosum	2	1	–	3	2	66.6
Taraxacum officinale	1	1	–	2	1	50.0
Trifolium repens	2	–	–	2	–	—

on July 23 and, from appearances, had been infected for some time as the leaves were badly spotted in many cases.

The conidia produced on the sugar beet are apparently capable of infecting many host plants, as shown by the results of inoculations recorded in table II. Infections were obtained on 26 species of plants in 17 genera of 12 families (fig. 5). These species were not confined to closely related genera or families (figs. 3 and 4). The species in the genera Beta, Chenopodium, Rumex and Spinacea have been previously reported, but 16 species comprising 13 genera in 10 families, are newly reported hosts.

There appeared to be but little association between the relationship of species and the percentage of infection secured in greenhouse trials. *Chenopodium album* was much more readily infected than *Amaranthus retroflexus,* although both are closely related to *Beta vulgaris.* On the other hand, *Malva rotundifolia, Soja max, Ipomoea batatis, Apium graveolens, Plantago lanceolata, P. major, Arctium lappa* and *Lactuca sativa* were all

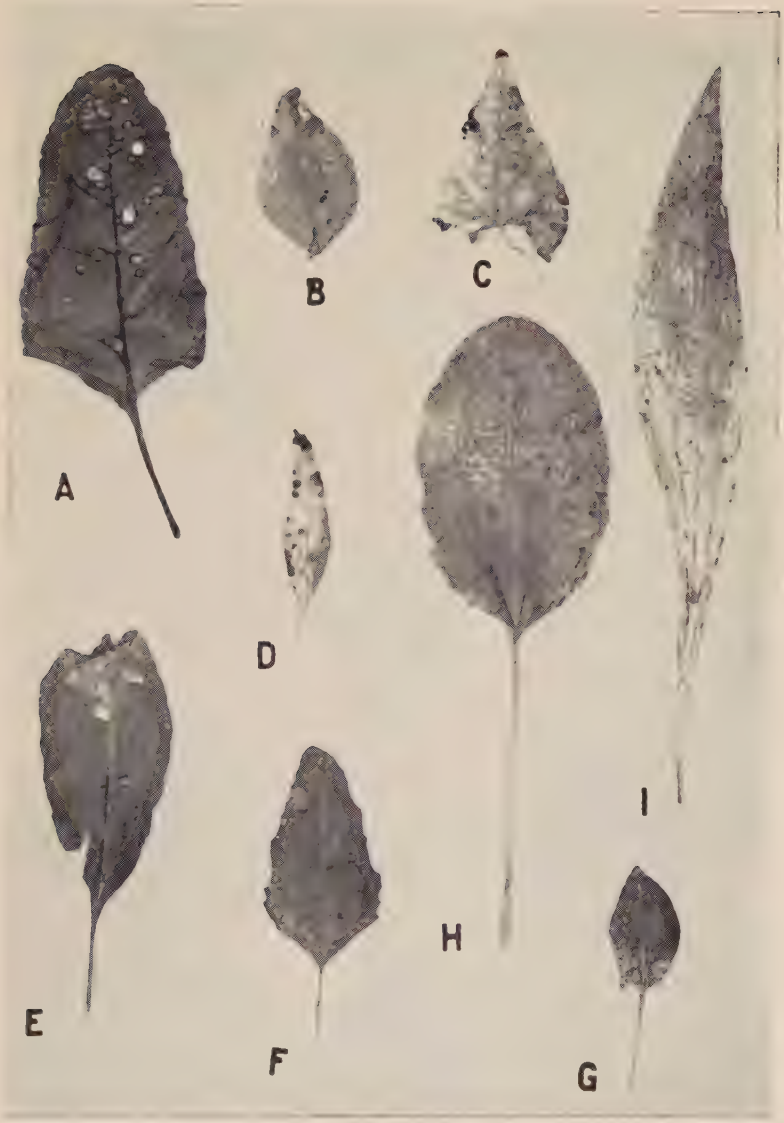

Fig. 3. Cercospora leaf-spot infection on leaves of nine species of plants artificially infected in the greenhouse with pure culture suspensions of *Cercospora beticola;* A. *Beta vulgaris;* B. *Amaranthus retroflexus;* C. *Polygonum convolvulus;* D. *Polygonum pennsylvanicum;* E. *Rumex crispus;* F. *Chenopodium album;* G. *Rumex acetosella;* H. *Plantago major;* I. *Plantago lanceolata.*

relatively easily infected, although not closely related to Beta. Within the genus the ease of infection was equally variable. *Polygonum convolvulus* was readily infected, while *P. pennsylvanicum* was infected with difficulty. Among the legumes in-

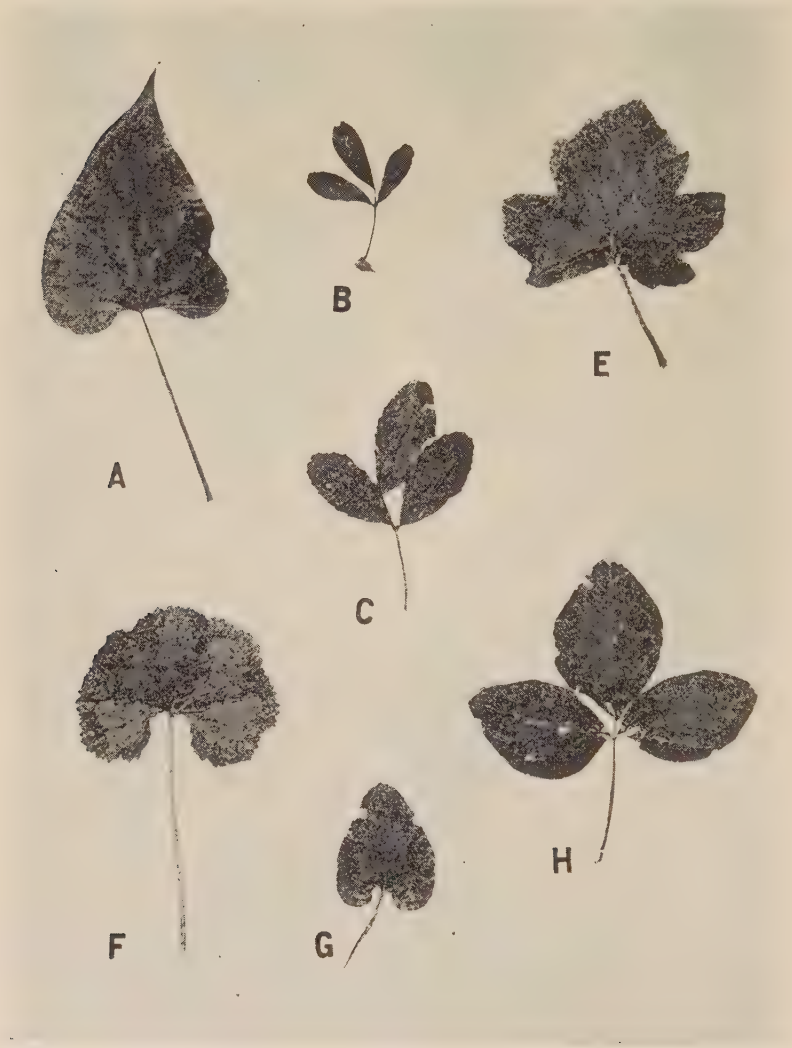

Fig. 4. Typical leaf-spot infection on leaves of seven species of plants artificially inoculated in the greenhouse with pure culture suspensions of *Cercospora beticola*: A. *Ipomoea batatas;* B. *Medicago sativa;* C. *Melilotus alba;* E. *Citrullus vulgaris;* F. *Malva rotundifolia;* G. *Arctium lappa;* H. *Soja max.*

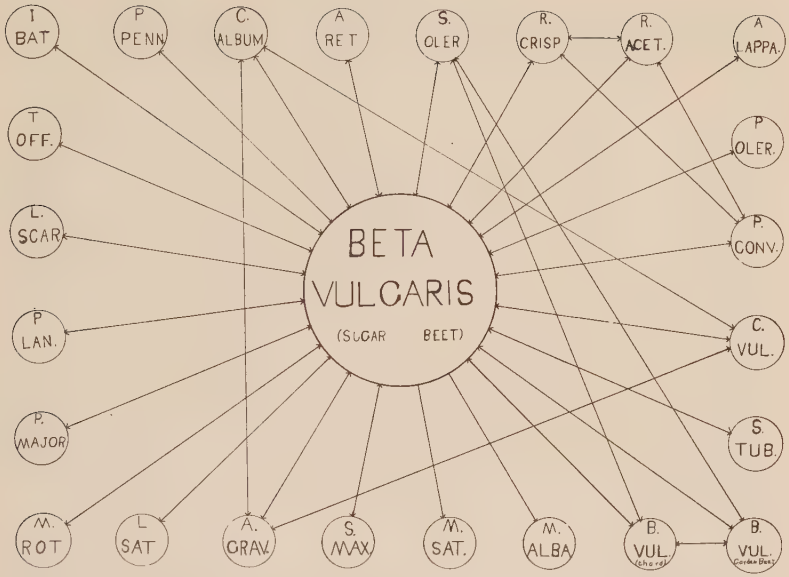

Fig. 5. Diagram indicating infections obtained upon a number of plants by *Cercospora beticola* isolated from *Beta vulgaris*. Arrows indicate the direction of the inoculation; when pointed they indicate positive inoculations.

oculated only *Soja max* appeared readily infected. *Medicago lupulina, Trifolium repens* and *Pisum sativum* were not infected in any of the trials, while *Medicago sativa* and *Melilotus alba* were only slightly infected.

An examination of the infected plants revealed that in many cases the fungus was growing on the old dead and dying leaves. In some cases these leaves were in an advanced state of necrosis; this was particularly true of *Lactuca sativa, Solanum tuberosum* and *Taraxacum officinale*. This would indicate that under certain conditions the fungus may live as a saprophyte upon the old dead leaves other than those of sugar beet. Lettuce, growing beside some heavily infected garden beets in a garden at Ames, Iowa, was found to have many dead leaves around the base. When these dead leaves were taken into the laboratory and placed in a moist chamber, typical *Cercospora beticola* conidia and conidiophores were produced. When suspensions of conidia of the isolation from the above mentioned lettuce leaves were sown upon sugar beet leaves, they produced infection as readily as did the isolation from sugar beet.

At the same time that the inoculation studies with *C. beticola* were made upon the various host plants listed in table II, microscopic measurements of the conidia and conidiophores were made. These measurements consisted of the average length, width of

TABLE III. CONIDIA AND CONIDIOPHORE MEASUREMENTS OF *CERCOSPORA BETICOLA*, TAKEN FROM THE VARIOUS HOST PLANTS AFTER ARTIFICIAL INOCULATION IN THE GREENHOUSE WITH PURE CULTURES.

Plant inoculated	Conidia				Conidiophores			
	Length μ	Width		Average number septations	Length μ	Width		Average number septations
		Apical end μ	Basal end μ			Apical end μ	Basal end μ	
Amaranthus retroflexus	146.8	2.2	5.2	6.1	94.3	5.38	6.0	3.1
Apium graveolens	205.1	1.8	5.0	15.68	188.2	5.8	5.8	4.28
Arctium lappa	177.0	1.61	5.4	16.28	67.7	5.52	6.0	1.6
Beta vulgaris (sugar beet)	219.6	2.36	5.96	22.32	79.2	4.24	5.96	2.96
Beta vulgaris (garden beet)	173.0	2.08	4.56	19.0	89.5	4.48	4.68	2.6
Beta vulgaris (Swiss chard)	196.0	2.20	4.71	20.4	84.4	4.3	4.52	2.7
Chenopodium album	228.0	2.08	5.16	26.8	157.0	4.08	4.41	4.35
Citrullus vulgaris	207.5	2.44	5.6	15.8	165.0	4.4	5.88	3.92
Ipomoea batatis	182.0	1.8	5.33	18.5	94.5	4.07	4.2	2.6
Lactuca sativa	204.0	2.56	6.12	22.56	167.8	5.6	5.66	5.2
Lactuca scariola	175.4	2.6	5.75	20.5	144.0	6.05	6.0	5.05
Malva rotundifolia	183.4	2.04	5.6	19.3	161.5	4.7	5.07	4.8
Medicago sativa	—	—	—	—	—	—	—	—
Melilotus alba	78.16	3.24	5.62	6.6	63.28	6.4	6.8	2.08
Melilotus officinalis	—	—	—	—	—	—	—	—
Plantago lanceolata	195.2	2.4	5.4	21.4	92.4	4.3	4.7	2.9
Plantago major	216.5	2.35	5.65	24.8	105.8	4.25	4.5	2.5
Portulaca oleracea	177.1	2.04	4.8	15.7	79.8	5.2	5.4	1.88
Polygonum convolvulus	181.1	2.48	6.08	24.4	96.4	4.1	4.4	2.4
Polygonum pennsylvanicum	102.88	2.04	4.44	9.0	80.68	5.56	6.31	1.91
Rumex acetosella	223.9	2.32	5.12	19.5	102.0	4.5	5.0	2.4
Rumex crispus	185.0	2.88	5.7	16.72	92.7	4.7	6.2	2.9
Soja max	154.5	2.14	5.16	22.6	92.9	5.6	5.8	2.6
Solanum tuberosum	192.4	2.21	5.3	20.0	86.6	5.1	5.31	3.1
Spinacea oleracea	202.3	1.96	4.84	24.24	113.0	5.2	5.12	4.0
Taraxacum officinale	193.2	2.04	6.36	21.9	122.6	5.3	5.7	2.88

apical and basal ends and number of septations of 25 conidia or conidiophores chosen at random. The results of the measurements are tabulated in table III. A comparison of these figures will show a wide variation between the measurements of the conidia and conidiophores taken from the various hosts inoculated with *C. beticola.*

Conidia lengths varied from 78.16μ to 228.0μ. The average conidial length for the 26 different host plants was 162.06μ. The isolation in each instance was incubated in the laboratory in the same moist chamber so that the measurements represent the growth under similar conditions. Their morphology, however, might not be comparable with that of the conidia produced under field conditions.

RESULTS OF CROSS INOCULATIONS

Whenever one of the host plants mentioned in table II became infected with *Cercospora beticola,* the fungus was isolated and

carried in pure culture. Suspensions of conidia from these cultures were sprayed on the host from which the culture was isolated to test the pathogenicity under greenhouse conditions. The results obtained are recorded in table IV.

Due to the limited time after the discovery of the infected host plant, some of the isolations were used much less than others. In all cases the first host chosen included the species from which the isolation was made, the sugar beet, and, where possible, species closely related to the original host.

CERCOSPORA BETICOLA FOUND IN THE FIELD

From time to time plants in the vicinity of Ames and Kanawha, Iowa, were found infected with a species of Cercospora. The fungus was isolated and attempts made to reinfect the same species and to inoculate sugar beets as well as other plants. Six plants found in the field, namely, *Amaranthus retroflexus, Chenopodium album, Lactuca sativa, Malva rotundifolia, Melilotus alba* and *Polygonum convolvulus,* were infected with a Cercospora which appeared morphologically and pathologically similar to *Cercospora beticola.* The names of the plants and the results of the inoculation trials are shown in table V. An examination of the results will show that sugar beet plants were readily infected by all of the isolations (fig. 6).

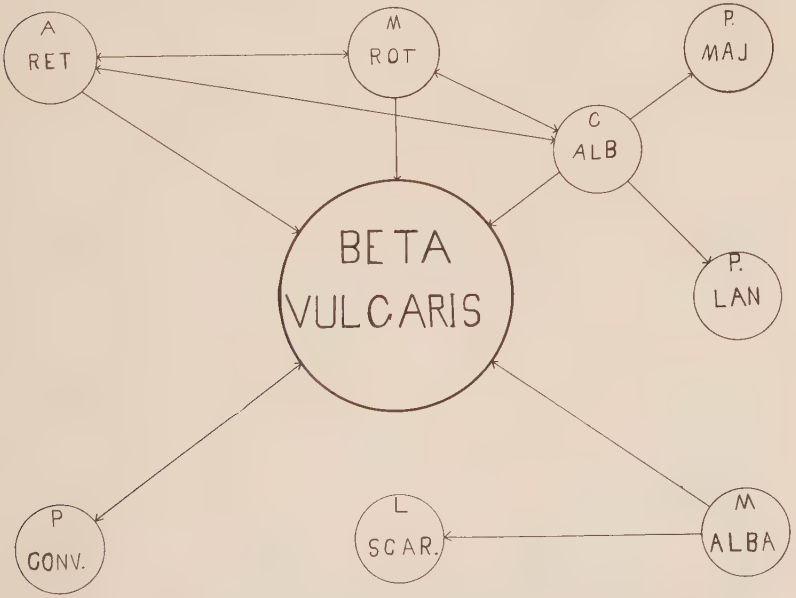

Fig. 6. Species of Cercospora from various hosts which infected *Beta vulgaris.* Arrows indicate the direction of the inoculations; when pointed they indicate positive inoculations.

TABLE IV. CROSS INOCULATION OF *CERCOSPORA BETICOLA* ON 24 DIFFERENT SPECIES OF PLANTS.

Plants inoculated	Hosts from which isolations were made																											
	Amaranthus retroflexus		*Arctium lappa*		*Beta vulgaris*		*Chenopodium album*		*Citrullus vulgaris*		*Ipomoea batatis*		*Lactuca sativa*		*Malva rotundifolia*		*Plantago lanceolata*		*Plantago major*		*Polygonum convolvulus*		*Polygonum pennsylvanicum*		*Portulaca oleracea*		*Soja max*		*Spinacea oleracea*	
	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.
Amaranthus retroflexus	1	0			1	10	1	0																						
Apium graveolens					5	2																								
Arctium lappa			2	2	6	0																								
Beta vulgaris (sugar beet)	4	0	4	0			15	0	11	0	4	0	6	0	4	0	4	0	3	0	12	0	4	0	4	0	4	0	8	0
Beta vulgaris (Swiss chard)					9	0	6	0															12	0						
Beta vulgaris (garden beet)					12	0	6	0			1	0					4	0	1	0	12	0							4	0
Chenopodium album	1	0			11	5	5	9	0	1	1	0							0	1	1	5							1	3
Citrullus vulgaris					14	10			12	4																			1	1
Ipomoea batatis					5	1					2	0	1	0					1	0										
Lactuca sativa					4	4			5	0			2	1																
Lactuca scariola					1	1																								
Malva rotundifolia	1	0			8	0	1	0							3	1	1	0												
Medicago lupulina					0	2																								
Medicago sativa					1	2																					0	2		
Melilotus alba					1	3																					0	2		
Pastinaca sativa					0	3							0	1			3	0												
Plantago lanceolata					5	1	1	0									3	1	1	1	0	3								
Plantago major					6	2	1	0	1	0			0	1			2	2	2	0	1	2								
Polygonum convolvulus					7	1	4	2	1	0			1	0					1	0	8	0	1	1						
Polygonum pennsylvanicum					1	6															1	5	0	2						
Portulaca oleracea					1	2													1	0					1	3				
Rumex acetosella					5	6	2	1													3	0								
Rumex crispus					7	9	2	1													4	2								
Soja max					5	1	1	2	1	0			1	0					1	0							2	2		
Spinacea oleracea					12	0	6	0					1	0			4	0	1	0	12	0							4	0
Solanum tuberosum					2	1																								
Taraxacum officinale					1	1																								

Cercospora on *Amaranthus retroflexus* was collected among some sugar beets growing on the Ash avenue experimental plots at Ames, Iowa. The plants showed numerous spots on the yellow fallen leaves which, upon incubation in a moist chamber for 24 hours, produced typical *Cercospora beticola* conidia. Many of the leaves were infected with white rust (*Cystopus candidus* Pers. Lev.), and Cercospora was found growing on about 40 percent of these necrotic acreas produced by the Phycomycete. In this instance Cercospora behaved like a saprophyte. When this fungus was isolated and inoculated upon the plants listed in table V, positive results were secured in each case comparable with those secured with *Cercospora beticola* from sugar beets.

The species of Chenopodium did not appear to be readily susseptible to infection with *Cercospora beticola.* As previously stated, Pool and McKay (36) and Schmidt (41) failed to secure infection on *Chenopodium album.* It had been previously infected artificially, however, in the greenhouse by Nagel (31). When an isolation of a *Cercospora sp.* from old fallen leaves of *Chenopodium album* collected at Kanawha, July 10, 1932, was sprayed over the host plants listed in table V, positive results were obtained in every case. Old fallen leaves of *Chenopodium album* collected in the garden of Dr. J. C. Gilman, Ames, Iowa, on July 24, 1932, produced typical *Cercospora beticola* conidia when placed in the moist chamber.

TABLE V. RESULTS OF INOCULATIONS UNDER GREENHOUSE CONDITIONS WITH PURE CULTURES OF A *CERCOSPORA SP.* ISOLATED FROM NATURALLY INFECTED PLANTS COLLECTED IN THE FIELD

Host plant inoculated	Hosts from which isolations were made											
	Amaranthus retroflexus		*Chenopodium album*		*Lactuca sativa*		*Malva rotundifolia*		*Melilotus alba*		*Polygonum convolvulus*	
	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.	Pos.	Neg.
Amaranthus retroflexus	1	0	1	0								
Beta vulgaris (sugar beet)	4	0	4	0	4	0	4	0	2	0	3	0
Chenopodium album	1	0	2	0								
Malva rotundifolia	1	0	2	0			2	0				
Medicago sativa									0	2		
Melilotus alba									0	2		
Melilotus officinalis									0	1		
Plantago lanceolata	1	0	1	0								
Plantago major			1	0								
Polygonum convolvulus											1	1

Over 50 percent of the attempts to infect *Chenopodium album* artificially with *Cercospora beticola* were successful and the isolation from these affected plants infected 12 other plants as shown in table IV. This, coupled with the fact that *Chenopodium album* collected from two widely separated places in the state bore a Cercospora which appeared identical with *Cercospora beticola,* indicates that the weed may be a factor in the spread of this leaf-spot upon sugar beets in the field.

Plants of *Malva rotundifolia* growing wild at Ames were found to be infected with a Cercospora which readily infected *Beta vulgaris* and *Malva rotundifolia* under greenhouse conditions. Table VI shows that the length and septation number of conidia and conidiophores from the plants grown in the vacant lot were much less than for the average produced in the greenhouse (table III), but they are within the range of *Cercospora beticola* measurements.

Melilotus alba collected at Kanawha, Iowa, July 10, 1932, proved to be infected with *Cercospora sp.* Pure cultures of this fungus infected *Beta vulgaris,* but in the greenhouse trials failed to infect *Medicago sativa, Melilotus alba* or *Melilotus officinalis.* Morphologic measurements were somewhat less for this isolation than for the others secured, but they are within the range of *Cercospora beticola.*

Cercospora sp. isolated from some partly dead *Polygonum convolvulus* leaves collected at Kanawha, July 10, 1932, infected

TABLE VI. CONIDIA AND CONIDIOPHORE MEASUREMENTS OF A *CERCOSPORA SP.* ISOLATED FROM NATURALLY-INFECTED PLANTS COLLECTED IN THE FIELD.

Host, and location	Conidia				Conidiophores			
	Length μ	Width		Average number septations	Length μ	Width		Average number septations
		Apical end μ	Basal end μ			Apical end μ	Basal end μ	
Amaranthus retroflexus (Ash avenue, Ames)	233.	2.06	5.6	20.5	112.4	5.6	5.6	3.7
Chenopodium album (Kanawha, Iowa)	218.	2.51	5.41	24.56	134.0	6.0	6.4	4.73
(Garden, Dr. J. C. Gilman, Ames)	216.	2.21	5.32	25.6	140.0	5.0	5.5	4.40
Lactuca sativa (Garden, Dr. Gilman)	204.	2.56	6.12	22.56	167.8	5.6	5.66	5.2
Malva rotundifolia (North end, greenhouse, Ames)	77.6	3.7	4.9	8.01	70.68	5.04	6.8	2.68
Medicago sativa (North end, greenhouse, Ames)	123.3	2.88	6.2	9.6	87.0	5.72	5.76	2.84
Melilotus alba (Kanawha, Iowa)	67.2	3.33	6.1	4.88	90.5	5.57	5.88	2.3
Polygonum convolvulus (Kanawha, Iowa)	195.5	2.3	6.0	22.25	156.3	6.1	6.4	5.8

both *Beta vulgaris* and *Polygonum convolvulus* when plants of these two species were inoculated with pure cultures in the greenhouse. Conidia and conidiophore measurements were typical for those of *Cercospora beticola* produced under similar conditions.

On Aug. 16, 1932, a species of Cercospora was found on leaves of *Polygonum convolvulus* collected a short distance from the place of collection of July 10 mentioned above. The morphologic characters were typical of *Cercospora beticola*, but the necessity for closing the season's work reported here prevented isolation and study of pathogenicity.

Table VI is a tabulation of the measurements of the *Cercospora sp.* isolated from the host plants collected in the field. Although there is a wide range of measurements shown, it is not greater than that shown by *Cercospora beticola* when artificially inoculated on to the same host plants under artificial conditions, as shown in table III.

CONTROL OF CERCOSPORA LEAF-SPOT

Efforts toward the control of Cercospora leaf-spot of sugar beets were directed along two major lines; first, through the use of fungicides and, second, by the modification of cultural practices. The use of fungicides embodied the common practice of keeping the foliage covered with a protective coat to prevent infection by the fungus; whereas the modification of cultural practice was concerned with the reduction of humidity among the beets.

SPRAYING AND DUSTING FOR CERCOSPORA LEAF-SPOT

In 1899 and again in 1909 Duggar (16, 18) suggested bordeaux for the control of Cercospora leaf-spot in the United States. In 1914, Townsend (45) reported satisfactory control by this method.

European workers, however, have given more attention to control by means of fungicides than investigators in this country. Costa (11), Griessing (22), Kresl (25), Sedlmayr (43), Saillard (40), de la Loma (14) and Schmidt (41) all report encouraging results from the use of bordeaux. The increase in yield of beets and sugar in the sprayed fields more than paid for the material and labor.

Copper lime dusts have also been given extensive trials both in America and in Europe. In Italy, Gabotto (20), Pratolongo (37) and Mori (30) report that the returns from the use of copper-lime dusts more than paid for the material and labor used in the application.

In America, Coons, Stewart and Larmer (10) have given copper-lime dust extensive trials. They used a power duster at

Rocky Ford, Colo., and report that three applications of 35 pounds per acre, costing from $6 to $8, produced sufficient increase in yield to more than pay the cost of application.

SPRAYING AND DUSTING IN 1929

During the season of 1929, the value of fungicides as a control for leaf-spot was tested under field conditions at Mason City, Iowa. Bordeaux mixture, copper hydroxide paste manufactured by the Roessler Hasslacher Company, New York City, and copper-lime dust, known as the Niagara Dri-Copper, manufactured by the Niagara Sprayer Company of Middleport, N. Y., were the fungicides used. Of these only the results obtained with the copper-lime dust will be reported here.

The dust was applied by means of a Niagara Sprayer Company 12-row power duster equipped to hold 12½ pounds of copper sulfate and 50 pounds of hydrated lime at one mixing, which was sufficient to cover 2 acres of sugar beets. All dust was applied during the early morning hours, while the beet leaves were covered with dew and complete coverage could be secured.

All data for sugar percentage and purity were based on five 5-beet samples collected at random from each of the various plots and analyzed in the laboratory of the American Beet Sugar Company at Mason City, Iowa. Yield data were secured either by selecting five representative 5-beet samples from each plot, or by obtaining actual yields from the weigh slips received by the growers at the receiving dumps.

On a 50-acre farm near Britt, Iowa, plots which extended the length of the field were established 24 rows wide. The dusted plots alternated with the undusted plots, the yields from the middle 12 rows only being considered. Beginning on Aug. 9, the dusted plots were covered every 10 days. Plot 1 received five applications of copper-lime dust, while plot 3 received only three applications (table VII). Plots 2 and 4, used as checks, were not dusted. Each plot consisted of approximately 1¼ acres. An examination of the data will show that the dusted plots reveal a very slight increase in the percentage of sugar, whereas check plot 2 actually yielded more pounds of beets per acre than plot 1 which received five applications of the dust.

TABLE VII. INFLUENCE OF MONOHYDRATE COPPER SULFATE UPON THE YIELD AND SUGAR PERCENTAGE OF SUGAR BEETS WITH *CERCOSPORA BETICOLA*.

Plot number	Number dusts	Percentage sugar	Yield per acre—lbs.
1	5	15.19	13,297
2	0	15.17	13,474
3	3	14.85	15,515
4	0	14.71	15,345

Fig. 7. At left, a sugar beet badly diseased with *Cercospora beticola,* showing circle of dead leaves typical of leaf-spot injury. At right, a healthy plant of sugar beet.

On two neighboring fields, plots were established and treated as the ones in the 50-acre field described above. In one of these fields leaf-spot failed to develop and the yields in the fall failed to show any differences between treated and untreated plots. In the other field, a deep tillage experiment which crossed the dusted plots was carried on and rendered the results unreliable.

On a field in which leaf-spot failed to develop by harvest time, but in which dusted plots had been given three applications, samples were secured, weighed for yield and analyzed for sugar percentage and purity. The object in taking these samples was to determine to what extent copper-lime dust might influence the composition of the beets. Samples were secured from three of the dusted and three of the undusted plots. The results are recorded in table VIII. It will be seen that the differences which appeared are probably not significant.

TABLE VIII. INFLUENCE OF MONOHYDRATE COPPER SULFATE ON THE WEIGHT, PURITY AND SUGAR CONTENT OF BEETS FREE FROM CERCOSPORA LEAF-SPOT.

Plot no.	Dusted plots			Check plots		
	Average weight per beet in oz.	Average purity	Average percentage sugar	Average weight per beet in oz.	Average purity	Average percentage sugar
1	19.75	87.1	16.72	18.25	88.9	17.27
2	15.25	86.7	17.60	19.50	89.1	16.72
3	19.00	90.0	16.65	19.25	84.8	16.07

SPRAYING AND DUSTING IN 1930

During 1930 spraying and dusting were again carried on near Britt, Iowa. The series of plots was more extensive than in 1929. In the largest field the sugar beet webworm attacked the

Fig. 8A. A field of sugar beets badly infected with Cercospora leaf-spot.

sugar beets, badly damaging them and rendering any results unreliable. In two other fields leaf-spot failed to develop in epidemic form and the results secured at harvest time failed to show either positive or negative effects of the dust.

THE INFLUENCE OF SPACING UPON THE DEVELOPMENT OF OF CERCOSPORA LEAF-SPOT

Biologic and pathologic studies reviewed on preceding pages suggested that Cercospora leaf-spot might be inhibited by modification of existing cultural practices. The custom of blocking and thinning provides favorable conditions for the development and spread of Cercospora leaf-spot. Beets spaced 12 inches in the row produce a dense mat of overlapping foliage, resulting in a high relative humidity. This situation favors the development of Cercospora, as shown by Pool and McKay (36), who pointed out the necessity of a relatively high humidity in the relationship between stomatal opening and infection. They found that it was only after the beet leaves became large enough to shade the ground that infection took place.

The question of proper spacing distance for best sugar beet yields has been given a great deal of attention in Europe during the past few years but little regard was given to the effect of such spacing on development of sugar beet diseases. Chmelar and Mikolasek (2, 3, 4) and Chmelar and Simon (5, 6, 7) reported results of previous experiments in which spacing 45x30 cm. and 45x25 cm. (equivalent to 18x12 inches and 18x10 inches) gave the best yields. These authors, who have carried on the most extensive investigations, also reported their own experiments from 1922 to 1927, using various spacings and types of beets. They concluded that under the conditions of their experiments the distance between plants in the row is of

Fig. 8B. A neighboring field of healthy sugar beets.

more importance than the distance between rows. The distance between rows is regulated by the cultivating machinery. This is not true of the distance between beets in the row.

Davies (13) carried on experiments in England in 1927, 1928 and 1929 which favored even closer spacing between the rows as well as between the beets in the row. His best yields were with rows from 12 to 16 inches apart and the beets not more than 10 inches apart in the row.

Melhus and Aikman (29) reported that in 1930 the cross cultivated beets (22x20 inches) outyielded the normally thinned beets (22x12 inches) 1,962.92 pounds per acre. During the same year, Vestal and Bell (46) measured the difference in humidity between variously-spaced beets and found that when beets in 22-inch rows were spaced 12 inches apart the humidity stood at 100 percent, an average of 2.3 hours longer each night than the humidity in similar rows spaced 20 inches apart. It is believed this difference in the time exposed to a saturated atmosphere was largely responsible for the fact that there was six times as much infection in the 12-inch spacing as in the 20-inch.

FIELD TRIAL IN 1931

The work reported by Melhus and Aikman (29) and Vestal and Bell (46) showing that wide spacing retards the development of Cercospora leaf-spot was studied further in 1931.

The field used for the experiments in 1931 at Kanawha contained a fraction over 14 acres, 12 of which were plotted in the experiment itself. From north to south the field was 290 feet and from east to west, 2,100 feet. From north to south the field was measured off into ten 29-foot areas running the length of the field. The entire field was planted to sugar beets. The row widths in the various 29-foot areas were 18, 20, 22, 24, 26, 24, 22, 20, 18 and 16 inches.

When the beets were large enough for blocking, the field was measured from east to west into 30-foot areas. The first area extended from north to south across all of the planted rows and was hand-blocked and thinned to 12 inches in the row. The second, third and fourth areas were blocked by means of a Planet Jr. four-row cultivator so that when thinned the beets were 20 inches apart in the rows in the second area, 24 in the third and 20 in the fourth. This series was duplicated 12 times across the field with this difference: Each alternate series was 12, 24, 20, 24-inch instead of the order given in the first series. This scheme made possible a combination of the 12, 20 and 24-inch blockings with all of the various row widths. All of the beets were cultivated three times in the direction in which they were planted and the 24 and 20-inch blockings were cross cultivated twice.

ARTIFICIAL INOCULATION

The inoculation plan included plots in each of the spacing combinations outlined above. For this work an area was selected which, because it was on low ground, appeared to be most likely to develop an epidemic of leaf-spot. The four rows nearest the checks in each of the blockings were selected for treatment and harvest. To insure an epidemic, artificial inoculations were made on Aug. 3, 12 and 19. All inoculations were made with finely powdered diseased leaf material dusted uniformly over the leaves of the beets in the late evening after the dew had gathered. The data were taken from the two middle rows.

The infection counts of Cercospora leaf-spot were based on the amount of infection per leaf (table IX). A leaf which was not infected was listed as "disease-free." A leaf which had fewer than 50 spots was considered as "lightly infected." A leaf which had 50 or more spots on it was considered as "heavily infected," and any leaf which was no longer green but yellow and brown was considered "dead." All of the leaves upon which infection counts were made were included under one of the above classes.

DEVELOPMENT OF CERCOSPORA LEAF-SPOT IN THE DRILLED AND CHECKED BEETS

The first leaf-spot was found among the plots in the 12-inch blocking in the 18-inch rows on Aug. 1. The attack first appeared along the north edge of the field and spread southward. A heavy rain fell on July 30 and 31, sufficient to give the infection a start. By Aug. 4 the infection was spreading among all the 12-inch blockings to the extent of about 10 percent. The first leaf-spot was found among the checked beets Aug. 10.

Another heavy rain fell Aug. 17, and by Aug. 19 infection was general over the field.

On Aug. 28 and 29 and Sept. 29 and 30, readings were made on 24 consecutive plants in each rate of thinning (table IX). At the time of the first reading, the 20 and 24-inch blockings contained the largest number of lightly infected leaves, while the 12-inch blocking contained about five times as many heavily infected leaves. There were also more dead leaves among the beets in the 12-inch blocking than among those of the 20 and 24-inch blockings. The disease-free leaves were less variable for all blockings for both readings.

The percentage of infected leaves at the later reading, however, shows an interesting change. The percentages of light infection among the beets of all blockings were approximately the same. The number of heavily infected leaves on the beets in the 12-inch blockings had decreased at the later reading through the death of many of those listed as heavily infected since the first reading. The heavily infected leaves on the beets in the 20 and 24-inch blockings had increased since the first reading, and, while there had been an increase in the number of dead leaves, the increase was much less than found on the beets in the 12-inch blocking. These data show that the infection appeared first on the beets in the 12-inch blocking and did not appear on the beets of the wider blocking until about 10 days later.

Results were secured by averaging the data from representative (table X) 1/1,000 acre plots selected from each row width. The yield of beets in tons per acre increased as the width of rows increased up to 20 and 22 inches; thereafter the yield decreased. As previously stated, the infection development among the beets in the 20 and 22-inch blockings was about 10 days later than that among those in the 12-inch blocking. This 10-day delay in the epidemic among the beets of the wider blockings meant just that much longer use of all the photosynthetic area. The older leaves were retained for a longer period by the beets in the 20 and 24-inch blockings than by those in the 12-inch. This permitted a longer period of photosynthetic activity with a corresponding increase in yield.

An analysis of variance by the method of Fisher (19) showed significant differences in the yields of both rows and blocks. The differences between adjacent blockings are significant; those between adjacent row widths are not. The data in table X indicate that the 20-inch blocking is superior to the others tried when combined with 18 to 22-inch row widths.

An analysis of the pounds of sugar per acre in the beets seemed to parallel the yields of beets.

Within the limits of the data of this report, there is no evi-

TABLE IX. COMPARISON OF CERCOSPORA LEAF-SPOT INFECTION OF SUGAR BEET LEAVES IN 12, 20 AND 24-INCH ROWS AT KANAWHA, IOWA, 1931.

Blocking in inches	Row width in inches	Date initial infection	Leaf infection, Aug. 28-29						Leaf infection, Sept. 29-30					
			Percentage disease free	Percentage light infection	Percentage heavy infection	Percentage dead	Relative humidity		Percentage disease free	Percentage light infection	Percentage heavy infection	Percentage dead	Relative humidity	
							Day av.	Night av.					Day av.	Night av.
12	16	Aug. 1*	25.5	19.5	36.8	18.2			25.5	5.9	14.9	53.7		
	18		24.5	16.3	41.9	17.3			25.8	4.8	15.4	54.0		
	20		22.8	20.1	43.5	13.6	82	93	26.9	4.8	12.9	55.4	89	99
	22		26.6	26.4	30.6	14.4			28.2	4.8	16.8	50.2		
	24		26.8	27.8	26.6	18.8			27.9	4.6	16.9	50.6		
	26		27.2	35.1	21.8	15.9			27.9	5.2	25.3	41.8		
20	16	Aug. 10	26.0	42.4	18.3	13.3			24.5	4.9	23.3	47.3		
	18		31.8	56.2	2.5	9.5			27.4	5.8	30.3	36.5		
	20		26.4	51.8	10.9	10.9	76	88	24.1	3.1	24.4	38.3	83	96
	22		30.9	56.8	3.9	8.4			29.6	6.8	29.6	34.0		
	24		28.5	57.9	3.1	10.5			27.3	5.8	38.4	28.5		
	26		26.6	59.9	5.9	7.6			29.2	8.1	33.9	28.8		
24	16	Aug. 10	35.4	50.4	8.8	5.4			34.3	4.9	23.4	37.4		
	18		31.0	57.0	3.6	9.2			29.5	5.8	29.7	35.0		
	20		30.4	46.4	13.2	10.0			28.9	6.1	24.7	40.3		
	22		29.6	54.8	7.7	7.8			30.9	5.8	28.4	34.9		
	24		26.3	59.2	4.7	9.8			32.5	6.8	28.4	32.3		
	26		29.1	58.2	2.3	10.4			30.3	7.2	25.7	36.8		

*Ten percent of leaves infected by Aug. 4. Infection general by Aug. 19.

TABLE X. THE YIELD IN TONNAGE, SUGAR AND PURITY PER ACRE OF THE 12, 20 AND 24-INCH BLOCKINGS IN THE 16, 18, 20, 22, 24 AND 26-INCH ROW WIDTHS AT KANAWHA, IOWA, IN 1930 AND 1931.

Row width in inches	12-inch blocking				20-inch blocking				24-inch blocking			
	Tons beets per acre	Percentage sugar	Percentage purity	Lbs. sugar per acre	Tons beets per acre	Percentage sugar	Percentage purity	Lbs. sugar per acre	Tons beets per acre	Percentage sugar	Percentage purity	Lbs. sugar per acre
16	12.6	13.2	77.1	3,024	13.3	13.2	81.4	3,498	12.4	12.6	79.4	2,929
18	13.8	13.6	83.0	3,740	14.9	13.5	81.5	4,017	13.8	14.1	80.2	3,866
20	14.0	14.8	82.8	4,144	15.0	13.9	81.9	4,170	14.5	14.4	82.8	4,190
22	13.5	14.8	82.2	3,996	15.4	13.9	83.1	4,281	13.3	14.1	81.4	3,758
24	11.7	15.1	83.3	3,443	14.5	13.7	80.6	3,975	12.8	13.9	80.9	3,548
26	11.1	13.9	81.8	3,085	14.2	14.2	82.3	4,047	12.2	13.2	81.6	3,300

dence that the blocking widths produced any significant effect upon the percentage of sugar, although the percentage in the 12-inch blocking was greater than that in the 20 and 24-inch blockings. The percentages in the different row widths were significantly different, with the greatest percentage found in the 20-inch rows.

No significant differences in the purity were found. Such small differences as were encountered indicate that increasing the blocking distance does not reduce the purity. The greatest average percentage, again, is found in the 20-inch row widths.

The sugar percentages and yields obtained from the 12 and 20-inch blockings for the years 1930 and 1931 are shown in table XI. It will be seen that while the sugar percentage was higher for the 12-inch blocking the total amount of sugar secured was greater for the 20-inch blocking. This is explained by reference to table X, which shows the per acre yield of the 22-inch rows to be 15.4 tons for the 20-inch blocking and 13.5 tons for the 12-inch blocking.

TABLE XI. A COMPARISON OF THE 1930 AND 1931 SUGAR, PURITY AND YIELD OF BEETS IN 12 AND 20-INCH BLOCKINGS, 22-INCH ROWS.

	1930		1931	
	12-inch blocking	20-inch blocking	12-inch blocking	20-inch blocking
Percentage sugar	15.61	14.56	14.80	13.90
Percentage purity	80.50	82.50	82.15	83.10
Yield in beets in tons per acre	14.40	15.53	13.50	15.40
Yield of sugar in pounds per acre	4,508.0	4,528.0	3,996.0	4,281.0

COMPARISON OF ENVIRONMENTAL FACTORS

At the time the infection studies were being conducted in the field, environmental factors were also being studied. Temperature measurements between the rows for the season showed a day average of 2° F. higher in the 20-inch blocking than among the 20-inch drilled beets, while the night temperature was on the average 2° F. lower for the 20-inch than for the 12-inch drilled. The soil temperature in the 20-inch blocked beets showed a daily average of from 3 to 4° F. higher and a night temperature average of 5° F. lower than the 20-inch drilled beets. Light in the 20-inch blocked beets, as measured by the Clements' photometer, showed an average of two times the intensity of that in the 12-inch blocked beets. The wind, measured by the Tycos hand anemometer, showed in the 20-inch blocked beets two times the velocity of that in the 12-inch blocked. These factors resulted in a 10 percent higher evap-

oration rate among the 20-inch blocked as compared with the 12-inch blocked beets, when measured by means of Livingston porcelain atmometer cups set among the various row widths. The above data substantiate those secured by Vestal and Bell (46).

These factors all combined to reduce the humidity in the 20-inch blocked beets from 3 to 6 percent below that in the 12-inch blocked beets. The differences obtained in the daytime were slightly greater than those obtained at night. As previously stated in this article, differences of as much as 5 percent materially reduced the number of spores which germinated (table I). Thus the reduction in the amount of infection which occurred among beets in the 20-inch blocking at the time of the late readings as compared with that in the 12-inch blocking may have been due in part, at least, to the reduction of the humidity in the 20-inch blocking.

LITERATURE CITED

(1) Brown, W. On the germination and growth of fungi at various temperatures and in various concentrations of oxygen and CO_2. Ann. Bot. **36**:257-283. 1922.

(2) Chmelar, F., and Mikolasek, F. Vergleichende Versuche mit verschiedener Standweite der Rübenpflanzen im Mähren in den Jahren 1924 und 1925 und Ubersicht der Ergebnisse in den Jahren 1922 und 1923. Zeitschrift für die Zuckerindustrie der Cechoslovakischen Republik. **51**:697-706. 1927.

(3) ————————. **52**:325-327. 1928.

(4) ————————. **53**:177-180. 1929.

(5) Chmelar, F., and Simon, J. Vergleichende Versuche über verschiedene Zuckerrübenstandweite im Mähren im Jahre 1922. Zeitschrift für die Zuckerindustrie der Cechoslovakischen Republik. **47**:671-677, 683-694, 695-703, 707-711. 1922.

(6) ————————. **48**:309-316, 317-323. 1923.

(7) ————————. Ein Bekämpfungsversuch der *Cercospora beticola* in der Suranyer Zuckerfabrikswirtschaft im Jahre 1927. Zeitschrift für die Zuckerindustrie der Cechoslovakischen Republik. **52**:700-824. 1928.

(8) Coons, G. H. Sugar beet disease control progressing toward solution. U. S. Dept. Agr. Yearbook, **1928**:557-559. 1928.

(9) Coons, G. H., and Larmer, F. G. The physiology and variations of *Cercospora beticola* in pure culture. Mich. Acad. Sci. Papers. **11**:75-104. 1930.

(10) Coons, G. H., Stewart, D., and Larmer, F. G. The sugar beet leaf spot disease and its control by direct measures. U. S. Dept. Agr., Circ. 113. 1930.

(11) Costa, T. Contributo allo studio della *Cercospora beticola* Sacc. nella bassa vallata Padana. Nuovo Giornale Botanico Italiano. **34**:25-27. 1928.

(12) Cunningham, H. S. A study of the histologic changes induced

in leaves by certain leaf-spotting fungi. Phytopath. 23:717-751. 1928.

(13) Davies, W. Morley. The spacing of sugar beets. Great Britain Jour. Min. of Agr., 37:973-985. 1931.

(14) de la Loma, J. L. Cercospora in Spain. La Hacienda. 25:173-174. 1930. Original not seen. Abstracted in Facts about Sugar. 25:423. 1930.

(15) Duggar, B. M. Early blight of celery. New York (Cornell) Agr. Exp. Sta., Bul. 132:201-206. 1897.

(16) ————————. Three important diseases of the sugar beet. New York (Cornell) Agr. Exp. Sta., Bul. 163. 1899.

(17) ————————. Physiologic studies with reference to the germination of certain fungus spores. Bot. Gaz., 31:38, 66. 1901.

(18) ————————. Fungous diseases of plants. Ginn and Company, Boston. 1909.

(19) Fisher, R. A. Statistical methods for research workers. Oliver and Boyd, Edinburgh, fourth ed. 1932.

(20) Gabotto, L. La *Cercospora beticola* nel. Il Coltivatore, 1924. 71:108-112. 1925. Original not seen. Abstracted in Rev. App. Myc., 4:520. 1925.

(21) Gilman, Joseph C., and Archer, W. Andrew. The fungi of Iowa parasitic on plants. Iowa State Jour. of Sci., 3:299-507. 1929.

(22) Greissing, J., Secker, F., and Pollak, A. Ein Bekämpfungsversuch der *Cercospora beticola* in der Suranyer Zuckerfabrikswirtschaft im Jahre, 1927. Zeitschr. für Zuckerind., 52:325-327. 1928. Original not seen. Abstracted in Rev. App. Myc., 7:613. 1928.

(23) Hull, K. L. Studies in resistance and susceptibility of *Zea mays* L. to *Puccinia sorghi*, Schw. Physiologic Form 1. Chicago Univ. Abstracts of Theses. Science, 7:317-319. 1928-29.

(24) Killian, C. Etudes comparatives des caractères culturaux et biologiques chez les Deutéromycétes et les Ascomycétes parasites. Ann. Sci. Nat. Bot. Ser., 10:101-292. 1928.

(25) Kresl, F. X., and Peska, V. A. Ein Beitrag zur Bekämpfung der *Cercospora beticola* auf Zuckerrübe. Zeitschrift für Zuckerindustrie der Cechoslovakischen Republik. 53:177-180. 1928. Original not seen. Abstracted in Rev. App. Myc., 8:152. 1929.

(26) Lieneman, Catherine. Host index to the North American species of the genus *Cercospora*. Missouri Botanical Garden Annals, 16:1-32. 1929.

(27) Lund, Carrol T. Cross cultivation. The Sugar Beet, 7:13-15. 1930.

(28) Melhus, I. E. Culturing of parasitic fungi on the living host. Phytopath. 2:197-205. 1912.

(29) Melhus, I. E., and Aikman, J. M. The comparative effect of drilled and cross cultivated beets on the spread of leaf spot and yield in Iowa. The Sugar Beet, 7:16-21. 1930.

(30) Mori, G. Baroni, and Costa, T. Esperimenti di latta contro la Cercospora della barbabietola in Italia. L'Indus. Sacc. Ital., 21:311-319. 1928. Original not seen. Abstracted in Rev. App. Myc., 8:9. 1929.

(31) Nagel, C. M. The sporulation and host range of six species of Cercospora. Unpublished thesis. Library, Iowa State College, Ames, Iowa. 1932.

(32) Nagel, C. M., and Dietz, S. M. The sporulation of five species of Cercospora in pure culture. (Abstract.) Phytopath, 22:20. 1931.

(33) Pammel, L. H. Fungous diseases of sugar beet. Iowa Agr. Exp. Sta., Bul. 15:234-354. 1891.

(34) Pool, V. W., and McKay, M. B. Relation of stomatal movement to infection by *Cercospora beticola*. Jour. Agr. Res., 5:1011-1038. 1916.

(35) ————————. Climatic conditions as related to *Cercospora beticola*. Jour. Agr. Res., 6:21-60. 1916.

(36) ————————. Field studies of *Cercospora beticola*. Phytopath., 8:119-136. 1918.

(37) Pratolongo, A., and Allen, M. P. Sulla constituzion e sulla proprieta di alcuni antiporonosporici piu usati. Giorn. Chim. Ind. Appl., 11:3-7. 1928.

(38) Saccardo, P. A. Sylloge Fungorum, 4:431. 1896.

(39) ————————. Sylloge Fungorum, 22:1432. 1913.

(40) Saillard, E. Production de graines de betteraves et de sucre en 1927. Compt. Rend. Acad. d' Agric. de France, 14:522-539. 1928.

(41) Schmidt, E. W. Untersweiungen über die Cercospora Blattfleckenkrankheit der Zuckerrübe. Zeitschr. Parasitenk., 1:100-137. 1928.

(42) ————————. Cercospora disease of the sugar beet in Spain. Deutsche Zuckerindustrie, 57:446-448. 1932.

(43) Sedlmayr, C. T. Erfahrungen bei der Bekämpfung der Cercospora in Ungarn. Fortschr. der Landw., 4:416-418. 1929.

(44) Solheim, W. G. Morphological studies of the genus Cercospora. Ill. Biol. Monog., 12:1-55. 1930.

(45) Townsend, C. O. Leaf spot, a disease of the sugar beet. U. S. Dept. Agr., Farmers' Bul. 618. 1922.

(46) Vestal, E. F., and Bell, F. G. A preliminary study of some environmental factors on the spread of Cercospora leaf-spot and yield in checked and drilled sugar beets. Amer. Jour. Bot., 18:706-716. 1931.

(47) Vosbein, O. Machinelles Verhacken der Zuckerrüben in Deutschland. Mitt. Deut. Landw. Gesell., 47:165-166. 1932.

(48) Welles, C. G. Observations on taxonomic factors used in the genus Cercospora. Science, n.s. 59:216-218. 1924.

(49) ————————. Taxonomic studies on the genus Cercospora in the Philippine Islands. Amer. Jour. Bot., 12:195-218. 1925.

(50) Wenzel, A. Beiträge zur Kenntnis der Blattfleckenkrankheiten der Zuckerrübe. Phytopath. Zeitschr., 3:519-529. 1931.